

The present invention concerns the use of a recently discovered *Listeria* phage with specific, essential and relevant properties, which makes it particularly suitable for identifying and controlling *Listeria* contamination of dairy products, facilities and equipment.

5 In addition to the general scientific literature on the subject, there is also patent literature that teaches the utility of phages in general to control bacterial contaminations in food processing plants and in foodstuffs. See for example U.S. Patent No. 5,006,347 issued on April 9, 1991, U.S. Patent No. 4,851,240 issued on July 25, 1989, and EP 0414304A2 published on February 27, 1991. However, none of
10 the above discussed patents disclose a *Listeria* phage which was actually tested and shown to successfully control bacterial contamination in food processing plants and in food products. The reason for this is that all of the *Listeria* phages known in the art at the time of the disclosure in the previous patents were temperate phages, and were therefore not efficient at nor suitable for industrial bacterial eradication purposes. The
15 term "temperate" refers to the fact when a strain of phage injects its DNA into a bacterial target, the phage DNA integrates into the DNA of the host cell, as a "prophage", and can remain integrated therein for considerable periods of time. Since the prophage excises (and initiates replication and lysis) only when the host cell becomes stressed, the ensuing bacterial lysis is unpredictable and not easily
20 controlled, which is why temperate phages do not lend themselves well to industrial applications. Temperate phages are unsuitable for industrial decontamination purposes for other reasons as well, including the fact that they can deliver unwanted and dangerous genes to the bacteria target into which their DNA integrates. In contrast, there is a class of phages that lyse bacterial targets directly, given that they
25 do not have the molecular machinery required to integrate into the bacterial targets. Such phages are referred to as being "virulent" or "lytic" for the bacterial targets. Virulent phages against *Listeria monocytogenes* were discovered recently, by one of the present inventors.

The first of these virulent *Listeria* phages, designated A511, was described in
30 the literature in 1990 (see Loessner et. al., Applied and Environmental Microbiology, June 1990, p.1912-1918, 1990). The virulent phage according to the present invention belong to the Myoviridae family and have tails which contract towards the virus head. One particularly preferred phage is designated P100 and was deposited at the

We Claim:

1. A method for controlling *Listeria* contamination in a food product, on food processing equipment, or on food storage containers, comprising applying at least one strain of virulent phage selected from the group consisting of the Myoviridae family and virulent phage variants from the Siphoviridae family, to a food product or food processing equipment in an amount sufficient to reduce the amount of *Listeria*, wherein said lytic phage is virulent against *Listeria monocytogenes* strains of serovar 1/2.
2. The method according to claim 1, wherein said lytic phage is P100, ATCC patent deposit designation no. PTA-4383.
3. The method according to claim 2, wherein said P100 is applied in combination with phage A511, ATCC patent deposit designation no. PTA-4608.
4. The method according to claim 1, wherein said lytic phage is applied in combination with at least one agent selected from the group consisting of listeriolysin, a surface disinfectant, an antibiotic, a surfactant, an enzyme, and a phage specific for bacterial contaminants other than *Listeria monocytogenes*.
5. The method according to claim 1, wherein said food product is a dairy product.
6. The method according to claim 1, wherein said food product is an unpasteurized food product.
7. The method according to claim 1, wherein said food product is a meat product.
8. The method according to claim 6, wherein said meat product is a ready to eat meat product.
9. The method according to claim 1, wherein said food product is a fish product.
10. The method according to claim 1, wherein said food storage container is a salad bar and said food product is salad.
11. The method according to claim 1, wherein said food processing equipment is selected from the group consisting of a tube through which milk is being pumped, a high-salt content tank for processing cheese, a container from which cultures are applied to a surface of a cheese, a set of shelves on which a product is dried and cured, and a floor drain.

12. The method according to claim 1, wherein said lytic phage are applied by mixing with a liquid or semi-solid food product.
13. The method according to claim 1, wherein said lytic phage are mixed with a liquid and sprayed onto a surface selected from the group consisting of food products, food processing equipment and food storage containers.
14. The method according to claim 13 wherein said lytic phage are applied to said food processing equipment in combination with an agent selected from the group consisting of listeriolysin, a surface disinfectant, an antibiotic, a surfactant, an enzyme, and a phage specific for bacterial contaminants other than *Listeria monocytogenes*.
15. The method according to claim 1, wherein said lytic phage are lyophilized or cryopreserved by vitrification and applied in a dry form to said food product, food processing equipment and food containers.
16. A composition comprising phage P100, ATCC patent deposit designation number PTA-4383 and a carrier.
17. The composition according to claim 17, further comprising phage A511, ATCC patent deposit designation number PTA-4608.
18. The composition according to claim 17, further comprising an agent selected from the group consisting of listeriolysin, a surface disinfectant, an antibiotic, a surfactant, an enzyme, and a phage specific for bacterial contaminants other than *Listeria monocytogenes*.
19. The composition according to claim 16, wherein said carrier is a pharmaceutically acceptable carrier.
20. A method for treating an animal infected with *Listeria monocytogenes* comprising administering an amount of P100 suitable to reduce or eliminate said *Listeria monocytogenes*.
21. The method according to claim 20, further comprising administering phage A511.
22. Phage P100 deposited at the American Type Culture Collection, ATCC patent deposit designation number PTA-4383.
23. A method for detecting the presence of *Listeria monocytogenes*, comprising

obtaining a sample suspected to contain *Listeria monocytogenes*, incubating said sample with P100, and detecting any change in said sample caused by P100, as an indication of the presence of *Listeria monocytogenes*.

24. The method according to claim 23, wherein said change in said sample is due to lysis by P100 or a detectable label or signal.

25. The method according to claim 23, further comprising recombinantly inserting a gene construct into the genome of P100 before incubation with said sample, wherein expression of said gene construct results in a detectable signal in the presence of *Listeria monocytogenes*.

26. The method according to claim 25, wherein said gene construct encodes a bioluminescent protein.

27. The method according to claim 26 wherein said bioluminescent protein is selected from the group consisting of luciferase and a fluorescent protein.

28. The method according to claim 27, wherein said luciferase is from bacteria or insects.

29. The method according to claim 27, wherein said fluorescent protein is green fluorescent protein or a variant thereof.

30. The method according to claim 23, further comprising immobilizing said *Listeria monocytogenes* on a solid support and detecting any change on said solid support.

31. The method according to claim 30, wherein said *Listeria monocytogenes* are immobilized using anti-*Listeria* antibodies.

32. The method according to claim 31, wherein said solid support is a test strip.

33. The method according to claim 23, wherein said sample is obtained from a patient suspected of being infected with *Listeria monocytogenes*.
34. The method according to claim 23, wherein said sample is obtained from a food product, food processing equipment or food storage containers.
35. A purified endolysin protein obtainable from phage P100.
36. A method for controlling *Listeria* contamination in a food product, on food processing equipment or on food storage containers, comprising applying the endolysin protein according to claim 35, to a food product, food processing equipment or food storage container in an amount sufficient to reduce the amount of *Listeria*.
37. The method according to claim 36, further comprising applying at least one variety of lytic phage from the Myoviridae family to said food product, food processing equipment or food storage container.
38. The method according to claim 37, wherein said lytic phage is selected from the group consisting of P100 and A511.
39. The method according to claim 36, wherein said endolysin is recombinantly produced by another bacterial species.
40. The method according to claim 36, further comprising applying endolysin from at least one other phage which infects *Listeria* or another bacterial genera.
41. The method according to claim 40, wherein said other phage is A511.
42. The protein according to claim 35, wherein said endolysin protein is recombinantly produced.

43. A composition for controlling *Listeria* contamination in a food product, on food processing equipment or on food storage containers comprising endolysin protein obtainable from phage P100 and a suitable carrier.
44. The composition according to claim 43, further comprising at least one variety of lytic phage from the Myoviridae family.
45. The composition according to claim 44, wherein said lytic phage are selected from the group consisting of P100 and A511.
46. The composition according to claim 43, wherein said endolysin is recombinantly produced in a host bacteria.
47. The method according to claim 23, wherein a gene construct has been recombinantly inserted into P100 in order to provide or emit a signal confirming the detection of *Listeria monocytogenes*.
48. The method according to claim 47, wherein said gene construct is selected from the group consisting of genes encoding luciferase and green fluorescent protein.